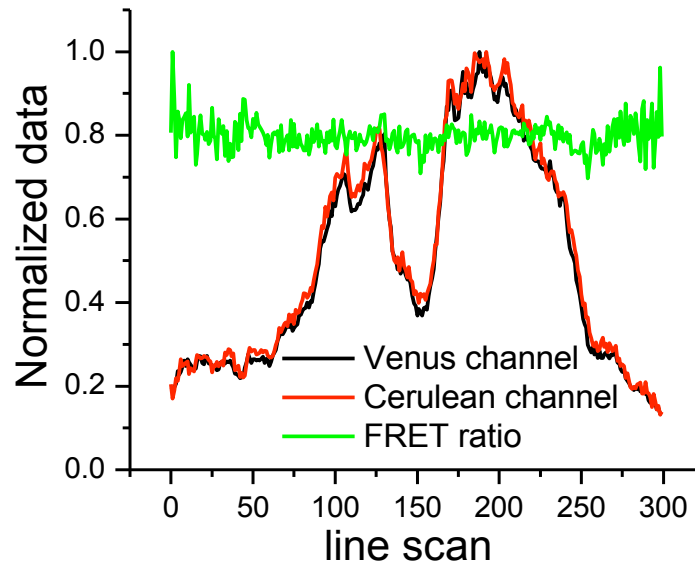
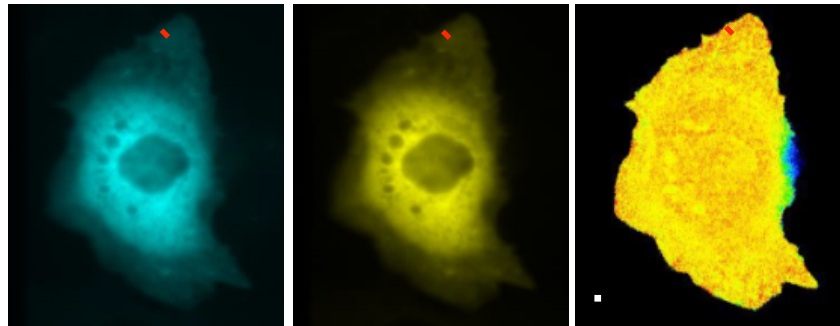


A



B



Supplementary Figure 3. FRET ratio is independent of concentration in actinin-sstFRET expressing cells. (A) Normalized line scans of donor channel, acceptor channel and FRET ratio image. Red and black trace shows 5 fold variation of fluorescence intensity increase across the cell in the acceptor and donor channels. The green trace indicates the FRET ratio is flat across the cell. (B) Images of cell expressing actinin-sstFRET. Acceptor channel, yellow; left panel, donor channel, cyan; middle panel and FRET ratio image, right panel. Red lines show the line scan over the images. Scale bar, 10 μm .